Benoxacor §82-1 Sunchronic Feeding in Mice

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DATA EVALUATION RECORD

Study Type: 13-Week Feeding Toxicity Study-Mice Guideline: §82-1 OPPTS 870.3100 (rodent)

DP Barcode: D223738
PC#: [redacted] SUBMISSION#: S446951
ID#: 7E03489

Test Material: Benoxacor Technical (purity 95%); CAS#: 41289-08-1

Synonym: CGA-154281

Dosages: 0, 50, 500, 2000 and 6000 ppm (0, 7.14, 70.7, 290 and 1100 mg/kg for males and 9.53, 99.8, 382 and 1470 mg/kg for females)

Citation: M. Bachmann (August 3, 1990) 3-Month Rangefinding Toxicity Study in Mice. Ciba-Geigy Ltd, 4332 Stein, Switzerland. Lab. Study# 891290. MRID#: 43337402, 43337403, and 43337404. Unpublished.

Sponsor: Ciba-Geigy Ltd, 4332 Stein, Switzerland

Background: This 13-week feeding toxicity study in mice consists of three reports. A final 13-week feeding toxicity study in mice (MRID#433374-01) plus an amendment (MRID#433374-03) and a supplement (MRID#433374-04) to the final report. The amendment and supplement only dealt with histopathological evaluations of the stomach tissues because of the concern of the stomach papilloma and carcinoma that were found in a chronic feeding study using Benoxacor in mice (MRID#428887-02). The two supplementary study reports are entitled, "Amendment to the Final Report: 3-Month Range-Finding Toxicity Study in Mice (Administered in Food) by M. Bachmann, March 14, 1994 (MRID#433374-03) and "Supplement to 3-Month Range Finding Toxicity Study in Mice. Re-evaluation of Stomach" by J.F. Hardisty, dated August 2, 1994 (Ciba-Geigy Ltd. Study#891290) (MRID#433374-04). This DER included evaluations of all three reports.

Executive Summary: Oral administration of benoxacor (96.8% pure) in mice via the diet at 50, 500, 2000 and 6000 ppm (≈ 7.14, 70.7, 290 and 1100 mg/kg/day for males and 9.53, 99.8, 382 and 1470 mg/kg/day for females) for 92-93 days produced the following major treatment-related effects:

Parameters		0 ppm	50 ppm	500 ppm	2000 ppm	6000 ppm
Body Weight	↓					♂
Water Consumption	↑				♂,♀	♂,♀
Hyperchromic & Macrocytic Anemia	↑					♂
White Blood Cell Count	↑					♀
Platelets	↑				♂,♀	♂,♀
Reticulocyte Count	↑					♂
Aspartate aminotransferase	↑					♂
Alkaline phosphatase	↑					♂
Liver weight	↑				♂,♀	♂,♀
Kidney weight	↑				♂,♀	♂,♀
Spleen weight	↑					♀
Liver necrosis	↑					♂,♀
Interhepatic bileduct hyperplasia	↑					♂,♀
Renal tubular lesion	↑					♂
Renal cortex fibrosis	↑				♂	♂
Renal cortex calcification	↑				♂	♂
Renal tubule atrophy	↑					♂

Based on the above data, the systemic toxicity NOEL is determined to be 500 ppm (70.7 and 99.8 mg/kg/day in males and females, respectively). The systemic toxicity LOEL is 2000 ppm (290 and 382 mg/kg/day in males and females, respectively), based on increased incidence of renal cortex fibrosis and calcification in males, and increased water consumption, increased platelet counts, and increased liver and kidney weights in both males and females.

Core Classification: **Acceptable-guideline.** This study satisfies the EPA's subdivision F guideline requirements (§82-1) for a 90-day Feeding Toxicity Study in mice.

Study Title: 3-Month Rangefinding Toxicity Study in Mice

A. OBJECTIVE

This 90-day feeding toxicity study was conducted to assess the range of systemic toxicity of Benoxacor following oral administration in the diet at 0, 50, 500, 2000 and 6000 ppm (0, 7.14, 70.7, 290 and 1100 mg/kg for males and 9.53, 99.8, 382 and 1470 mg/kg for females) dose levels.

B. MATERIALS AND METHODS

The in-life and necropsy phases as well as the histopathologic evaluations of this study were conducted at Ciba-Geigy Ltd, 4332 Stein, Switzerland.

Test Material

Physical Description: Solid (96.8% pure); Batch#: SG-7505-11
Storage Conditions: Room temperature in the dark.
Stability: Not provided in the study report

Test Animals

Species: Albino Mice (Tif: MAGf (SPF) hybrid NIH x MAG
Source: Animal Production, CIB-GEIGY Ltd, 4332 Stein, Switz.
Number of Animals: 100♂ and 100♀ ordered
Age: 4-5 weeks old at study initiation
Mean Starting Weights: ♂= 22.1 - 28.5 g; ♀= 21.0 - 24.8 g
Caging: Individually in Macrolon cage and soft wood bedding
Acclimation Period: 7 days
Feed: Pelleted Diet and water ad libitum.

Environmental Parameters

Ambient Temperature: 22±2°C; Relative Humidity: 55 ± 10 %;
Dark/Light Cycle: 12 hours; Room Air Exchanges: 16-20 per hour

C. STUDY DESIGN

Group Arrangement

Ten mice sex/group were used, housed individually, and acclimated for 7 days. Mice were randomly grouped as follows:

Groups	Dose Level ppm	No. of Animals Males/Females
Control	0	10/10
Low Dose	50	10/10
Mid Dose	500	10/10
High-Mid Dose	2000	10/10
High Dose	6000	10/10

Test Material Preparations and Analysis

The test compound was prepared by mixing the appropriate amount of benoxacor with the basal diet and about 25% water was added before pelleting. The pellets were then airdried. The amount of compound application was adjusted based on the most recent monthly body weight data. Homogeneity, Concentrations (94.8-98.5%) and the Stability (5 weeks) of the test article fell within acceptable ranges. The test material was administered orally in the diet (admixed to pelleted food). The controls were fed with similar pelleted food without the test article.

D. METHODS AND RESULTS

Clinical Observations

All rats were checked for mortality and clinical signs daily.

Results: Hunched posture and piloerection were noted in all ten 6000 ppm males. Endophthalmic eyes and labored breathing were noted in two and three 6000 ppm males, respectively (p. 30 of the study report). Three 6000 ppm males were found dead between days 6 and 8. Other deaths were due to trauma after blood collections (p. 30 of the study report).

Individual Body Weights and Food Consumption

Individual body weights were recorded prior to the start of the study, on the first day of dosing (day 0) and weekly thereafter. Food consumption was recorded weekly throughout the study.

Body Weights: Selected mean body weight and body weight gain data are as follows:

Table 1

Week	0 ppm ♂ / ♀	50 ppm ♂ / ♀	500 ppm ♂ / ♀	2000 ppm ♂ / ♀	6000 ppm ♂ / ♀
-1	25 / 23	25 / 23	26 / 23	25 / 23	25 / 24
1	26 / 22	27 / 22	26 / 22	25 / 22*	22**/ 21*
2	31 / 24	31 / 24	32 / 24	29 / 25	24**/ 24
4	33 / 25	33 / 25	34 / 26	32 / 26	27**/ 25
6	35 / 27	34 / 27	36 / 28	34 / 28	29* / 27
10	38 / 28	37 / 28	39 / 28	35 / 28	32* / 28
13	40 / 29	39 / 29	41 / 29	37 / 29	33* / 29
% BW gain Weeks -1 to 13	62.3% / 25.4%	58.8% / 26.6%	60.4% / 32.9%	45.8% / 25.8%	31.9% / 22.6%

* = $P < 0.05$; ** = $P < 0.01$; body weights were rounded off; from p. 32 of the study report; Body weight in grams.

The mean absolute body weight of the 6000 ppm males were statistically significantly reduced throughout the study period (22% less than the controls at termination). At termination, the body weight gain of the 6000 ppm males was significantly decreased as compared to the controls. This body weight reduction is judged to be treatment-related. The mean absolute body weights of the 2000 ppm males were slightly depressed (3.5-8.2% below the controls). In the 2000 and 6000 ppm females, minimal mean absolute body weight reductions were noted at week 1, but recovered to normal levels for the rest of the study period. The body weight gain was slightly decreased in the 6000 ppm females as compared to the controls.

Food Consumption: The mean absolute food consumption and food intake ratio (in shaded rows) data are as follows:

Table 2

Week	0 ppm ♂ / ♀	50 ppm ♂ / ♀	500 ppm ♂ / ♀	2000 ppm ♂ / ♀	6000 ppm ♂ / ♀
-1	38 / 32	35 / 33	37 / 34	36 / 34	37 / 35*
1	27 / 20	26 / 22	25 / 22*	16** / 19	7** / 20*
	144.7/124.7	136.7/139.8	135.3/143.0*	91.3**/123.4	44.7**/135.6
2	37 / 32	36 / 33	35 / 35*	38 / 34	47 / 51*
	170.8/188.6	169.4/194.5	160.9/205.7	189.2/197.0	274.7**/305.8*
4	39 / 36	38 / 37	40 / 40	37 / 40	40 / 48
	165.6/203.8	167.2/210.8	168.1*/220.7	168.5/219.3	209.5**/273.4
6	38 / 39	36 / 37	38 / 40	36 / 40	39 / 50
	154.0/206.3	151.0/198.8	151.3/208.8	153.0/206.0	190.4*/259.9
10	40 / 43	39 / 43	41 / 43	39 / 43	47 / 51
	153.0/221.7	152.6/220.0	148.8/219.2	158.7/217.8	214.2**/262.2
13	40 / 44	39 / 48	40 / 46	38 / 43	53 / 58
	143.8/219.0	144.8/236.1	138.9/225.0	150.8/212.4	231.8*/286.6

* = $P < 0.05$; ** = $P < 0.01$; absolute food consumption values (in grams) were rounded off and are tabulated in the non-shaded rows; the calculated food intake ratio values (g/kg/day) are tabulated in the shaded rows; from p. 37 & 41 of the study report.

In the first week of the study, the mean absolute food intake and the food intake ratios of the 2000 and 6000 ppm males were significantly reduced. The food intake ratios were reduced by 37% and 70% in the 2000 ppm and 6000 ppm males, respectively. The calculated food intake ratio of the 6000 ppm males were statistically significantly increased after week 1. Increased mean absolute food intake as well as increased food intake ratios noted in the 6000 ppm males at weeks 2 and 9-13 and in 6000 ppm females at weeks 2-13 were attributed to food spillage (p.37-43 of the study report).

Water Consumption

The mean water consumption of 2000 and 6000 ppm mice was increased throughout the study; consumption by the 2000 ppm males was increased by 19-64% from week 13 to termination and by 6000 ppm males by 12-65% throughout the study; values in the 2000 and 6000 ppm females were increased by 6-38% and 13-86%, respectively (p.45 of the study report).

Clinical Pathology Evaluations

Blood samples were drawn from the orbital sinus of all surviving mice before terminal necropsy for hematological and clinical chemistry evaluations. The checked (✓) hematology and clinical chemistry parameters were evaluated in this study as per required guidelines:

a. Hematology:

✓Erythrocyte count (RBC)
✓Hemoglobin (HB)
✓Hematocrit (HCT)
✓Mean Cell Volume (MCV)
✓Mean Cell Hemoglobin (MCH)
✓Differential WBC count
✓Total leukocyte count (WBC)
✓Platelet count
Prothrombin time
✓Thrombocyte Count

Results: Pertinent hematological data are tabulated below:

Table 3

Parameters	0 ppm ♂ / ♀	50 ppm ♂ / ♀	500 ppm ♂ / ♀	2000 ppm ♂ / ♀	6000 ppm ♂ / ♀
Erythrocyte Count (T/l)	11 / 10	11 / 10	11 / 10	11 / 10	10* / 10
Hemoglobin (mmol/l)	10 / 9	10 / 10	10 / 10	10 / 10	10 / 10*
Mean Cell Volume (fl)	43 / 45	44 / 44	44 / 44	43 / 45	45**/ 44
Mean Cell Hemoglobin (fl)	0.9 / 1.0	0.9 / 1.0	0.9 / 1.0	0.9 / 1.0	1.0**/ 1.0
White Blood Cell Counts(G/l)	2.8 / 1.8	2.1 / 1.4	1.9 / 2.0	1.4**/ 2.0	2.4 / 2.4*
Platelets (G/l)	1376 / 1105	1362 / 1171	1399 / 1188	1525*/ 1279*	1480 / 1382**
Reticulocytes (l)	0.017 / ne	ne / ne	ne / ne	ne / ne	0.034*/ ne

* = $P < 0.05$; ** = $P < 0.01$; fl = Femtoliter; G/l = Billion/liter; mmol/l = millimole/liter; T/l = 10^{12}/liter; l = liter; ne = not evaluated; values were rounded off; from p.48-54 of the study report.

The data on Table 3 suggest that there was a slight indication of hyperchromic and macrocytic anemia in the 6000 ppm males, associated with an increased number of reticulocytes. These are considered to be treatment-related. Higher numbers of white blood cells in 6000 ppm females, blood platelets in 2000 ppm males and females as well as 6000 ppm females; increased number of white blood cells in 6000 ppm females and platelets in 2000 and 6000 ppm males and females are judged to be treatment-related (p.49-54 of

the study report).

b. Clinical Chemistry

√Total Bilirubin
√Creatinine
√Total Protein
√Aspartate aminotransferase (ASAT)
√Alanine aminotransferase (ALAT)
√Blood Urea Nitrogen
√Glucose
√Alkaline phosphatase
√Cholesterol
√Potassium
Chloride
Phosphorus
Calcium
√Sodium
√Albumin
√Albumin/Globulin ratio
√Triglycerides

Results: Pertinent clinical chemistry data are tabulated below:

Table 4

Parameters	0 ppm ♂ / ♀	50 ppm ♂ / ♀	500 ppm ♂ / ♀	2000 ppm ♂ / ♀	6000 ppm ♂ / ♀
Glucose (mmol/l)	6.5 / 5.4	6.9 / 5.8	7.5 / 6.2	7.7 / 5.8	7.8 / 7.9**
Urea (mmol/l)	11 / 7.6	10 / 9.1	9 / 8.1	10 / 7.4	9 / 7.0*
Creatinine (µmmol/l)	44 / 37	44 / 42	44 / 38	41 / 38	41* / 35*
Protein (g/l)	56 / 54	55 / 55	56 / 54	54 / 54	51* / 53
Albumen (g/l)	32 / 33	33 / 34	33 / 33	32 / 32	31* / 32
Globulin (g/l)	23 / 21	22 / 21	23 / 21	22*/ 22	21 / 21**
Albumen/Globulin Ratio (l)	1.4 / 1.6	1.5 / 1.6	1.4 / 1.6	1.5 / 1.5	1.5 / 1.6
Aspartate Aminotransferase(U/l)	58.5 / 107	76.8*/ 146	70.5 / 110	58.7 / 112	80.4**/ 122
Alanine Aminotransferase(U/l)	46.8 / 58.8	67.1 / 84.8	64.2 / 65.7	34.2 / 66.7	63.9 / 65.3
Alkaline phosphatase (U/l)	80.9 / 120	81.5 / 113	78.0 / 107	82.9 / 118	133.0**/ 120

* = P <0.05; ** = P <0.01; l = Liter; g/l = gram/liter; mmol/l= millimole/liter; U/l = International Unit/liter; ne = not evaluated; values were rounded off; from p.56-59 of the study report.

Increased aspartate aminotransferase and alkaline phosphatase were noted in the 6000 ppm males; these findings are related to treatment and consistent with significant changes affecting the liver discussed below (p. 56-59 of the study report). The albumen and globulin values were slightly decreased in 6000 ppm males and females.

Pathology:

All animals were sacrificed and necropsied at the end of the test period (days 91 and 92). The animals were anesthetized by ether and killed by exsanguination. All guideline-required organs, such as the brain, heart, liver, kidneys, spleen, adrenals,

testes, ovaries, spinal cord (cervical and thoracic), sciatic nerve, any gross lesions, and others were harvested and fixed in 10% neutral buffered formalin for further histopathological evaluation.

a. Gross Macroscopic Evaluations

The gross macroscopic examinations included physical examination of all external surfaces, all orifices, and all internal body cavities with their associated organs.

Results: Gross macroscopic evaluation did not reveal any treatment-related changes (p. 68-87 of the study report).

b. Organ Weights

All guideline-required organ weights: the brain, heart, liver, kidneys, adrenals, thymus, spleen, ovaries and testes, were recorded at terminal necropsy.

Results: Pertinent absolute and relative organ weights are tabulated below.

Table 5

Absolute and Relative Organ Weights	0 ppm ♂ / ♀	50 ppm ♂ / ♀	500 ppm ♂ / ♀	2000 ppm ♂ / ♀	60000 ppm ♂ / ♀
Terminal Body Weights (g)	37.6 / 28.1	38.1 / 28.7	39.0 / 29.2	33.6 / 28.4	30.6*/ 27.3
Brain Weight (g)	0.52 / 0.56	0.53 / 0.55	0.54 / 0.55	0.50 / 0.54	0.52 / 0.54
Brain/Body Weight Ratio	14.1 / 20.1	14.0 / 19.3	14.1 / 19.0	15.1 / 19.2	17.1*/ 19.7
Liver Weight (g)	1.95 / 1.62	2.07 / 1.77	2.20 / 1.83	2.18 / 1.89	2.37 / 2.20**
Liver Weight/Body Weight Ratio	52.0 / 57.8	54.5 / 61.6	56.3 / 62.8	64.5**/ 66.9*	77.3**/ 80.5**
Liver Weight/Brain Weight Ratio	376.7 / 288.2	392.4 / 320.6	409.5 / 333.7*	433.0 / 349.2**	453.1 / 410.6**
Kidney Weight (g)	0.54 / 0.41	0.55 / 0.42	0.59 / 0.44	0.56 / 0.47*	0.55 / 0.49**
Kidney Weight/Body Weight Ratio	14.4 / 14.7	14.5 / 14.7	15.3 / 15.1	16.7**/ 16.7*	17.9* / 18.1**
Kidney Weight/Brain Weight Ratio	103.7 / 73.3	104.8 / 76.5	110.2 / 80.0	111.9 / 87.2*	105.0 / 92.0**
Spleen Weight (g)	0.08 / 0.11	0.08 / 0.11	0.08 / 0.11	0.07 / 0.13	0.09 / 0.14*
Spleen Weight/Body Weight Ratio	2.07 / 3.85	2.08 / 3.78	1.93 / 3.88	1.95 / 4.48	2.82 / 5.02*
Spleen Weight/Brain Weight Ratio	14.8 / 19.1	15.0 / 19.7	13.9 / 20.7	13.0 / 23.3*	16.5 / 25.6**

* = P ≤0.05; ** = P ≤0.01; values were rounded off; from p.61-66 of the study report.

Based on the results from the organ weight data above, the following conclusions can be made:

Absolute mean liver weight increases showed dose-related trends in both sexes. Statistically significant increases of liver/body weight ratios were noted in the 2000 and 6000 ppm males and females, and they are judged to be treatment-related effects.

The absolute and relative mean kidney weights were statistically significantly increased in the females of the 2000 ppm (by 15% and 14%, respectively) and of the 6000 ppm (by 20% and 23%, respectively) dose levels. The absolute and relative mean spleen weights were increased in the 6000 ppm females and they are considered to be related to treatment (p.61-66 of the study report).

c. Histopathological Evaluations

All fixed tissues and organs of all dose groups as well as from found-dead and moribund-sacrificed animals were processed, sectioned, and stained with hematoxylin and eosin.

Results: Pertinent microscopic findings are as follows.

Table 6

Histopathological Findings	0 ppm	50 ppm	500 ppm	2000 ppm	6000 ppm
	10♂/10♀	10♂/10♀	10♂/10♀	10♂/10♀	10♂/10♀
Liver					
-Necrotic Hepatocytes	0 / 0	0 / 0	0 / 0	0 / 0	5 / 7
-Interhepatic Bile Duct Hyperplasia	0 / 0	0 / 0	0 / 0	0 / 0	3 / 8
Kidney					
-Acute Tubular Lesion	0 / -	0 / -	0 / 0	0 / 0	3 / -
-Renal Cortex Fibrosis	0 / -	0 / -	0 / -	2 / -	4 / -
-Renal Cortex Calcification	0 / 0	0 / 0	0 / 0	4 / 0	7 / 1
-Renal Tubule Atrophy	0 / -	0 / -	0 / -	0 / -	5 / -
-Renal Tubule Basophilic Proliferation	3 / 0	2 / 1	1 / 0	5 / 1	5 / 1

* = P ≤0.05; ** = P ≤0.01; - = not evaluated; derived from 88-97 of the study report.

Increased incidence of microscopic changes were noted in the 2000 and 6000 ppm groups. Increased incidence of interhepatic bile duct hyperplasia was noted in the 6000 ppm dose group, namely, in 3 of the 10 males and in 8 of the 10 females. Necrotic hepatocytes of the liver was noted in 5/10 males and 7/10 females of the 6000 ppm dose groups. The noted hepatic changes are considered to be related to treatment. Acute tubular lesions were seen in 3/10 of 6000 ppm males; renal cortex fibrosis occurred in 2/10 of the 2000 ppm and in 4/10 of the 6000 ppm males, respectively; renal cortex calcification was found in 4/10 and in 7/10 males of the 2000 ppm and 6000 ppm dose levels, respectively, and in 1/10 of the 6000 ppm females; renal tubule atrophy occurred in 5/10 of the 6000 ppm males; none of these microscopic findings were found in the controls. All renal changes in the 2000 and 6000 ppm males are judged to be treatment-related to treatment. All other findings are judged to be unrelated to treatment, because the incidence and/or severity observed in the treated groups were comparable to the controls.

d. Histopathological Re-evaluation of the Stomach

No histopathological changes of the stomach were noted in the original final report (MRID#433374-02). Microscopic re-evaluation of the original histopathological slides, in addition to 6 newly prepared sections, 3 from the paraplast-embedded and 3 from the formalin-fixed wet tissue of the stomach were presented in the amended report (MRID#433374-03) as follows:

Table 7. Re-evaluation of Microscopic Sections of Stomach Tissues

Histopathological Findings	0 ppm	50 ppm	500 ppm	2000 ppm	6000 ppm
	10♂/10♀	10♂/10♀	10♂/10♀	10♂/10♀	10♂/10♀
Nonglandular Stomach					
-Epidermal Cyst	0 / 1	0 / 0	1 / 0	0 / 0	0 / 0
-Inflammatory Cell Infiltration	5 / 3	2 / 1	2 / 2	1 / 1	1 / 1
-Lymphocytic Infiltration	0 / 0	1 / 1	0 / 0	0 / 0	0 / 0
-Epithelium Hyperplasia	1 / nr	0 / nr	0 / nr	1 / nr	0 / nr
Glandular Stomach					
-Inflammatory Cell Infiltration	1 / 1	2 / 3	3 / 4	0 / 1	1 / 2
-Lymphocytic Infiltration	0 / 0	0 / 1	0 / 0	1 / 0	0 / 0
Gastric Mucosa					
-Erosion	0 / 1	1 / 1	1 / 1	1 / 0	0 / 0
-Necrosis	1 / nr	0 / nr	0 / nr	1 / nr	0 / nr
-Ulceration	nr/ 0	nr/ 0	nr/ 1	nr/ 1	nr/ 0
-Hyperplasia	nr/ 1	nr/ 0	nr/ 0	nr/ 0	nr/ 0
Gastric Gland Dilatation	5 / 2	0 / 2	2 / 1	4 / 1	1 / 2

* = P ≤0.05; ** = P ≤0.01; nr = not reported; derived from 13-43 of the study report (MRID#433374-03).

The above data did not show any treatment-related changes in either the glandular or nonglandular stomach.
Jerry Hardisty of the Experimental Pathology Laboratories. Inc. (EPL) re-evaluated the slides prepared by Ciba-Geigy and presented his findings in a supplemental report (MRID#433374-04). EPL concluded that no treatment-related differences were observed in the nonglandular or glandular portions of the stomach of treated mice of either sex as compared to the controls. None of the changes observed in the nonglandular stomach were considered to be precursor lesions related to the development of tumors observed following chronic feeding of CGA154281 technical in mice.

E. CONCLUSIONS:

Oral administration of Benoxacor (96.8% pure) in mice via the diet at 50, 500, 2000 and 6000 ppm (≈ 7.14, 70.7, 290 and 1100 mg/kg/day for males and 9.53, 99.8, 382 and 1470 mg/kg/day for females) for 92-93 days produced the following major treatment-related effects:

Parameters		0 ppm	50 ppm	500 ppm	2000 ppm	6000 ppm
Body Weight	↓					♂
Water Consumption	↑				♂,♀	♂,♀
Hyperchromic & Macrocytic Anemia	↑					♂
White Blood Cell Count	↑					♀
Platelets	↑				♂,♀	♂,♀
Reticulocyte Count	↑					♂
Aspartate aminotransferase	↑					♂
Alkaline phosphatase	↑					♂
Liver weight	↑				♂,♀	♂,♀
Kidney weight	↑				♂,♀	♂,♀
Spleen weight	↑					♀
Liver necrosis	↑					♂,♀
Interhepatic bileduct hyperplasia	↑					♂,♀
Renal tubular lesion	↑					♂
Renal cortex fibrosis	↑				♂	♂
Renal cortex calcification	↑				♂	♂
Renal tubule atrophy	↑					♂

Based on the above data, the systemic toxicity NOEL is determined to be 500 ppm (70.7 and 99.8 mg/kg/day in males and females, respectively). The systemic toxicity LOEL is 2000 ppm (290 and 382 mg/kg/day in males and females, respectively), based on increased incidence of renal cortex fibrosis and calcification in males, and increased water consumption, increased platelet counts, and increased liver and kidney weights in both males and females.

Core Classification: **Acceptable-guideline.** This study satisfies the EPA's subdivision F guideline requirements (§82-1) for a 90-day Feeding Toxicity Study in mice.